

CYTOGENETIC STUDIES IN THE GENUS *ORNITHOGALUM* L.

II. KARYOTYPE ANALYSES IN SOME BIOTYPES OF *O. CONICUM* JACQ., *O. LACTEUM* JACQ., AND *O. THYRSOIDES* JACQ.¹.

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ABSTRACT

The karyotypes of twenty-four biotypes of *O. thyrsoides*, nine of *O. lacteum* and one of *O. conicum* were analysed. All the chromosomes could be identified on the basis of morphological characteristics. The species and biotypes investigated all had a diploid number of 12, the only exception being the *O. lacteum* forma nov. from Sutherland with $2n = 10$.

The latter karyotype, with its long metacentric chromosome bearing a satellite, was found to have resulted from a centric translocation between chromosomes II and III of the $2n = 12$ complement.

Although single polyploid plants were found in otherwise diploid populations, polyploidy played no evolutionary role in Leighton's *miniaturum-maculatum* group of *Ornithogalum* species.

The results indicated that bar one exception no distinction between taxa could be based upon karyological differences. These results are discussed in the light of the present concepts regarding biosystematics.

INTRODUCTION

The karyotype is defined as the chromosome complement of a species, and is further characterized by the number, form and size of the chromosomes. The karyotype was early recognised as a definite species character, which is reasonably constant in most individuals of a population; departures from the species pattern is attributed largely to recognisable aberrations (Swanson, 1957).

As pointed out by Stebbins (1950) and others, cytology became an accepted and exceedingly useful science in the hands of the taxonomist who was interested in something more than simple morphological criteria for defining species relationships. In fact, relationships within natural groups of species can scarcely be considered complete, in an evolutionary sense, without reliable cytotaxonomic information to reinforce conclusions based on morphological criteria.

Babcock (1942), Clausen (1940, 1951), Stebbins (1950), Turesson (1925) and others stressed the causal role of factors such as the degree of inbreeding, structure of the population, apomixis, number and form of the chromosomes, environmental variation, and introgressive hybridization, in the pattern of evolutionary divergence.

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Lewis (1955) revealed that the amount of gene exchange between individuals is dependant upon the number and form of the chromosomes, thus creating a more objective criterion for the grouping of individuals into taxonomic categories.

The aim of the current investigation was to collect detailed karyological data of some species and biotypes in the genus *Ornithogalum* L., to ascertain its usefulness in a biosystematic study. Special attention was paid to the chromosome number and morphology, such as the position of primary and secondary constrictions.

MATERIALS AND METHODS

The following species, forma (according to the classification of Leighton, 1944/45) and biotypes were investigated:

O. thyrsoides Jacq. biotypes from:

Bokbaai;	Citrusdal;	Clanwilliam;
Darling;	Durbanville;	Elsenburg;
Gordons Bay;	Jonkershoek;	Kirstenbosch;
Cape Town;	Kruisfontein, Knysna;	
Leipoldtville;	Malmesbury;	Melkbosstrand;
Porterville;	Paleisheuvel;	Riviersonderend;
Sandveld;	Stellenbosch;	Swellendam;
Welgevallen, Stellenbosch.		

O. thyrsoides Jacq. forma η biotypes from:

Calvinia,	Kamieskroon,	Springbok.
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O. thyrsoides Jacq. forma ϵ biotype from:

Grahamstown.

O. lacteum Jacq. biotypes from:

Aurora,	Darling,	Duikerfontein,
Clanwilliam,	Elandsbaai,	Graafwater,
Pakhuispas,	Vredendal.	

O. lacteum Jacq. forma nov. biotype from:

Sutherland.

O. lacteum Jacq. forma β (= *O. synanthifolium*) biotype from:

Grahamstown.

O. conicum Jacq. biotype from:

Nieuwoudtville.

The localities mentioned are indicated on the map in fig. 1.

The biotypes were all morphologically distinct even after being grown under the same environmental conditions.

The bulbs were allowed to root in damp soil, and the young root tips cut

at approximately 11.00 a.m. They were fixed and treated according to the schedule of Pienaar (1955).

The technique of Östergren and Heneen (1962) was also tried, but gave inferior results in the current material.

Idiograms were constructed after drawing the chromosomes at a magnification of $3,800 \times$ by means of a "Carl Zeiss" drawing apparatus, and the three parameters, described below, were taken into account.

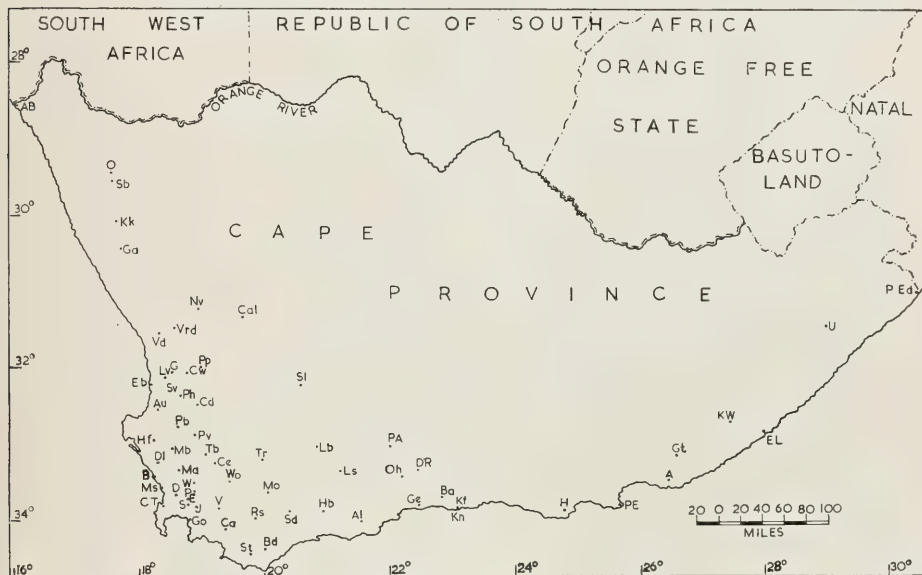
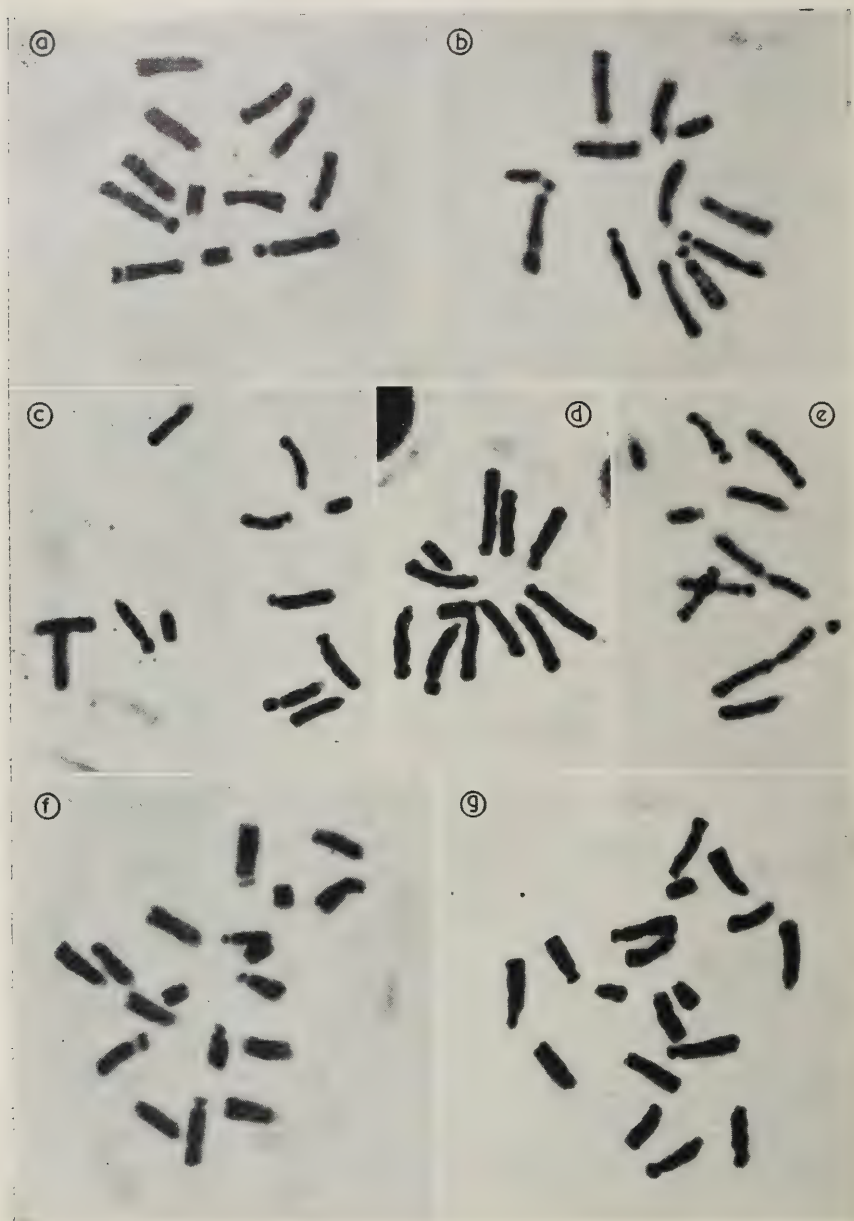


FIG. 1.

Map of the Cape Province indicating the localities where the various species and biotypes mentioned in the text were collected.

A—Alexandria; AB—Alexander Bay; Al—Albertinia; Au—Aurora; B—Bokbaai; Ba—Barrington; Bd—Bredasdorp; Ca—Caledon; Cal—Calvinia; Cd—Citrusdal; Ce—Ceres; CT—Cape Town; Cw—Clanwilliam; D—Durbanville; Dl—Darling; DR—De Rust; E—Elsenburg; Eb—Elandsbaai; EL—East London; G—Graafwater; Ga—Garies; Ge—George; Go—Gordons Bay; Gt—Grahamstown; H—Humansdorp; Hb—Heidelberg; Hf—Hopefield; J—Jonkershoek; Kf—Kruisfontein; Kk—Kamieskroon; Kn—Knysna; KW—King William's Town; Lb—Laingsburg; Ls—Ladismith; Lv—Leipoldtsville; Ma—Malmesbury; Mb—Moorreesburg; Mo—Montagu; Ms—Melkbosstrand; Nv—Nieuwoudtville; O—Okiep; Oh—Oudtshoorn; P—Paarl; PA—Prince Albert; Pb—Piketberg; PE—Port Elizabeth; PEd—Port Edward; Ph—Paleisheuwel; Pp—Pakhuispas; Pv—Porterville; Rs—Riviersonderend; S—Stellenbosch; Sb—Springbok; Sd—Swellendam; Sl—Sutherland; St—Stanford; Sv—Sandveld; Tb—Tulbagh; Tr—Touwsrivier; U—Umtata; V—Villiersdorp; Vd—Vredendal; Vrd—Vanrhynsdorp; W—Wellington; Wo—Worcester.



OBSERVATIONS

A total of 24 biotypes of *O. thyrsoides* Jacq. were investigated. All were found to have a diploid chromosome number of 12 (Fig. 2a — c; Fig. 3·1 — 3·24). Sporadic triploids were found in natural populations, e.g. one of a group of plants collected at a locality near Grahamstown (Fig. 2f).

Nine biotypes of *O. lacteum* Jacq. were similarly investigated and with only one exception, all proved to be normal diploids with $2n = 12$ (Fig. 2d; Fig. 3·25 — 3·33). Triploid plants also occur in natural populations as exemplified by a single plant, in a large collection from Pakhuispas, which had 18 somatic chromosomes (Fig. 2g).

The most interesting exception to the $2n = 12$ karyotype was found in the *O. lacteum* forma nov. biotype from Sutherland, with a chromosome number of $2n = 10$ (Fig. 2e; Fig. 3—33). This karyotype probably arose from a centric fusion (due to a translocation in the centromere region) between two long acrocentric chromosomes, since the length of the resultant metacentric chromosome was almost double that of the larger acrocentrics in the usual $2n = 12$ karyotype (see detailed explanation below).

O. conicum Jacq., like *O. thyrsoides* and *O. lacteum* had a somatic chromosome number of 12 (Fig. 3·34).

Idiograms of the haploid chromosome complements of the species and biotypes investigated, are presented in fig. 3.

Minor variations in chromosome lengths, arm ratios and secondary constrictions were detected, and in this respect a detailed study was undertaken in order to ascertain whether significant differences exist between biotypes and species.

In Table I the absolute lengths of chromosomes are given in μ .

DEDUCTIONS

From Table I, as well as from the idiograms in fig. 3, it is clear that the basic set is $x = 6$, and that it is fairly similar in the different species and biotypes. All the chromosomes are almost acrocentric (rod shaped) with slight differences in the length of the short arms. Chromosomes I, II, IV, V and VI can be dis-

FIG. 2.

(a-g) C=metaphase plates from root tip cells of some *Ornithogalum* species and biotypes: (a) *O. thyrsoides* from Elsenburg; (b) *O. thyrsoides* from Jonkershoek; (c) *O. thyrsoides* forma η from Kamieskroon; (d) *O. lacteum* from Elandsbaai; (e) *O. lacteum* forma nov. from Sutherland; (f) *O. thyrsoides* forma ϵ from Grahamstown, triploid plant; (g) *O. lacteum* from Pakhuispas, triploid plant. All figures $\times 2,000$.



FIG. 3.

Idiograms of the haploid chromosome complements of different *Ornithogalum* species and biotypes ($\times 3,000$)

1—20 *O. thyrsoides* Jacq. from:

- | | | |
|---------------------|-------------------|------------------|
| 1. Bokbaai | 2. Citrusdal | 3. Clanwilliam |
| 4. Darling | 5. Durbanville | 6. Elsenburg |
| 7. Gordons Bay | 8. Jonkershoek | 9. Kirstenbosch |
| 10. Kruisfontein | 11. Leipoldtville | 12. Malmesbury |
| 13. Melkbosstrand | 14. Paleisheuvel | 15. Porterville |
| 16. Riviersonderend | 17. Sandveld | 18. Stellenbosch |



19. Swellendam

20. Welgevalen

21—23 *O. thyrsoides* Jacq. forma η from:

21. Calvinia

22. Kamieskroon

23. Springbok

24. *O. thyrsoides* Jacq. forma ϵ from Grahamstown.

25—31. *O. lacteum* Jacq. from:

25. Aurora

26. Darling

27. Duikerfontein

28. Elandsbaai

29. Graafwater

30. Pakhuispas

31. Vredendal

32. *O. lacteum* Jacq. forma β (= *O. synanthifolium*) from Grahamstown.

33. *O. lacteum* Jacq. forma nov. from Sutherland.

34. *O. conicum* Jacq. from Nieuwoudtville.

TABLE I
AVERAGE LENGTHS OF CHROMOSOMES IN μ

Species and Localities	Chromosomes						Number of Cells
	I	II	III	IV	V	VI	
<i>O. thyrsoideis</i> , Jacq.							
Bokbaai	5.3	5.2	5.0	4.8	4.8	2.3	17
Citrusdal	5.5	6.0	5.2	5.8	6.1	2.6	10
Clanwilliam	5.5	5.3	5.5	5.2	4.8	2.2	9
Darling	5.6	5.5	5.3	5.4	5.3	2.4	10
Durbanville	5.9	5.5	5.5	5.5	4.9	2.5	11
Elsenburg	4.8	4.8	4.8	4.6	4.2	2.1	8
Gordons Bay	5.3	5.3	5.1	4.8	4.7	2.4	15
Jonkershoek	8.2	7.0	6.4	6.1	5.7	2.9	10
Kirstenbosch	5.1	5.0	5.4	4.7	4.9	2.4	9
Kruisfontein	5.9	5.9	5.6	5.6	4.8	2.5	8
Leipoldtville	4.8	4.8	4.6	4.8	4.2	2.1	6
Malmesbury	6.1	6.0	5.8	5.2	5.1	2.3	17
Melkbosstrand	5.8	5.7	5.8	5.5	4.9	2.3	10
Paleisheuvel	4.8	4.3	4.0	4.4	3.7	1.9	3
Porterville	5.8	5.8	5.8	5.7	5.6	2.5	10
Riviersonderend	5.6	5.6	5.5	5.2	5.2	2.6	8
Sandveld	5.6	5.3	5.4	5.1	4.9	2.4	10
Stellenbosch	6.6	6.3	5.9	5.9	5.5	2.7	10
Swellendam	6.1	6.0	6.3	5.7	5.6	2.6	10
Welgevallen	6.7	6.3	5.7	5.6	5.1	3.4	10
<i>O. thyrsoideis</i> , Jacq. forma η							
Calvinia	5.5	5.5	5.1	4.8	4.8	2.5	9
Kamieskroon	5.0	4.7	4.5	4.4	4.0	2.3	10
Springbok	5.1	4.9	4.7	4.6	4.5	2.3	12
<i>O. thyrsoideis</i> , Jacq. forma ϵ							
Grahamstown	5.9	5.5	5.3	5.2	4.2	2.6	10
<i>O. lacteum</i> , Jacq.							
Aurora	6.6	5.9	5.9	5.3	5.0	3.0	10
Darling	6.3	5.7	5.2	4.8	4.1	2.6	7
Duikerfontein	6.0	5.2	5.0	5.1	4.5	2.7	10
Elandsbaai	6.6	5.9	6.0	5.6	5.3	3.0	10
Graafwater	6.5	6.1	5.5	5.3	5.0	2.8	10
Pakhuispas	5.6	5.1	4.5	4.6	4.4	2.6	5
Vredendal	5.6	5.1	5.1	4.8	4.3	2.6	8
<i>O. lacteum</i> , Jacq. forma β							
Grahamstown	7.0	6.2	5.9	5.6	5.5	3.4	10
<i>O. lacteum</i> Jacq. forma nov.							
Sutherland	5.5	—	9.7	4.9	4.2	2.2	10
<i>O. conicum</i> , Jacq.							
Nieuwoudtville	5.3	4.8	4.6	4.4	4.1	2.2	6

tinguished by means of differences in arm ratios while chromosome III, which is similar in length to chromosome II, is characterized by having a satellite. Chromosome VI is very short and can easily be recognised.

It is evident from Table I that the Jonkershoek biotype has considerably longer chromosomes than any of the other biotypes and is almost a third longer

than those from Elsenburg and Leipoldville. This phenomenon is probably due to genetic differences, as in *Lolium perenne* (Thomas, 1936), *Lathyrus odoratus* (Upcott, 1937) and *Matthiola incana* (Lesley and Frost, 1927), where recessive mutants were reported to influence the spiralization of the chromosomes—a less compact spiralization producing a larger chromosome. In Table II it can be seen that the relative length of the Jonkershoek biotype corresponds closely to that of all the other biotypes, indicating that the difference in length is a regular feature of the whole chromosome complement.

Because the chromosomes of the C-metaphase plates of different cells in the same root tip exhibit variable degrees of contraction, and furthermore, due to presumed genetic differences, the chromosomes of the various biotypes are contracted to different degrees, the relative chromosome lengths (explained below) are generally considered more reliable for comparative purposes than absolute chromosome lengths.

Due to the absence of readily detectable differences between three of the chromosomes, the following parameters, as defined by Ford *et al.* (1960), were investigated to aid in the placement of the chromosomes in the idiograms.

A: The length of each chromosome relative to the total length of a normal haploid set, expressed as a percentage.

B: The arm ratio of the chromosomes expressed as the length of the longer arm relative to the shorter one.

C: The centromere index, expressed as the ratio of the length of the shorter arm to the whole length of the chromosome per 100.

Ford (1960) and his co-workers (A Human Chromosomes Study Group) used these parameters in describing the human karyotype. In Table II these parameters with corresponding letters are represented for all the different species and biotypes.

Table II proved very useful when comparing the different chromosomes of a complement, and the complements of the different biotypes, forma and species. It is obvious that little variation exists between the respective chromosomes of the different biotypes, forma and species. During evolutionary divergence the chromosomes of this group of species remained fairly constant. Of the small amount of karyotypic variation that does exist, there is as much between biotypes of a species as there is between species.

By means of Table II it was possible to determine which chromosomes were involved in the centric fusion, resulting in a metacentric chromosome, during the evolution of the $2n = 10$ karyotype of the *O. lacteum* forma nov. from Sutherland.

TABLE II

QUANTITATIVE PROPERTIES OF MITOTIC CHROMOSOMES IN *ORNITHOGALUM* L. (SEE TEXT FOR THE MEANING OF SYMBOLS A, B AND C).

SPECIES AND LOCALITIES			CHROMOSOMES		
I	A	19 5-5 15	19 27-3 4	19 15-5 6	19 15-5 6
	B	19 7-8 11	19 15-9 6	19 18-8 8	19 15-9 6
	C	19 8-4 11	19 13-3 13	19 16-3 13	19 13-3 13
	..	19 6-6 11	19 12-0 11	19 12-9 11	19 12-0 11
	..	19 6-4 11	20 12-9 11	20 12-9 11	20 12-9 11
	..	19 6-9 11	19 17-3 13	19 17-3 13	19 17-3 13
	..	20 6-4 11	20 12-9 11	20 12-9 11	20 12-9 11
	..	19 6-4 11	19 12-9 11	19 12-9 11	19 12-9 11
	..	19 6-4 11	19 12-9 11	19 12-9 11	19 12-9 11
	..	19 6-4 11	19 12-9 11	19 12-9 11	19 12-9 11
II	A	19 15-5 6	19 27-3 4	19 15-5 6	19 15-5 6
	B	19 7-8 11	19 15-9 6	19 18-8 8	19 15-9 6
	C	19 8-4 11	19 13-3 13	19 16-3 13	19 13-3 13
	..	19 6-6 11	19 12-0 11	19 12-9 11	19 12-0 11
	..	19 6-4 11	20 12-9 11	20 12-9 11	20 12-9 11
	..	19 6-9 11	19 17-3 13	19 17-3 13	19 17-3 13
	..	20 6-4 11	20 12-9 11	20 12-9 11	20 12-9 11
	..	19 6-4 11	19 12-9 11	19 12-9 11	19 12-9 11
	..	19 6-4 11	19 12-9 11	19 12-9 11	19 12-9 11
	..	19 6-4 11	19 12-9 11	19 12-9 11	19 12-9 11
III	A	18 13-6 7	18 13-6 7	18 13-6 7	18 13-6 7
	B	18 12-2 8	20 12-2 8	20 12-2 8	20 12-2 8
	C	18 12-2 8	20 12-2 8	20 12-2 8	20 12-2 8
	..	18 12-2 8	20 12-2 8	20 12-2 8	20 12-2 8
	..	18 12-2 8	20 12-2 8	20 12-2 8	20 12-2 8
	..	18 12-2 8	20 12-2 8	20 12-2 8	20 12-2 8
	..	18 12-2 8	20 12-2 8	20 12-2 8	20 12-2 8
	..	18 12-2 8	20 12-2 8	20 12-2 8	20 12-2 8
	..	18 12-2 8	20 12-2 8	20 12-2 8	20 12-2 8
	..	18 12-2 8	20 12-2 8	20 12-2 8	20 12-2 8
IV	A	17 15-6 6	17 15-6 6	17 15-6 6	17 15-6 6
	B	17 15-6 6	17 15-6 6	17 15-6 6	17 15-6 6
	C	17 15-6 6	17 15-6 6	17 15-6 6	17 15-6 6
	..	17 15-6 6	17 15-6 6	17 15-6 6	17 15-6 6
	..	17 15-6 6	17 15-6 6	17 15-6 6	17 15-6 6
	..	17 15-6 6	17 15-6 6	17 15-6 6	17 15-6 6
	..	17 15-6 6	17 15-6 6	17 15-6 6	17 15-6 6
	..	17 15-6 6	17 15-6 6	17 15-6 6	17 15-6 6
	..	17 15-6 6	17 15-6 6	17 15-6 6	17 15-6 6
	..	17 15-6 6	17 15-6 6	17 15-6 6	17 15-6 6
V	A	17 7-4 12	17 7-4 12	17 7-4 12	17 7-4 12
	B	17 7-4 12	17 7-4 12	17 7-4 12	17 7-4 12
	C	17 7-4 12	17 7-4 12	17 7-4 12	17 7-4 12
	..	17 7-4 12	17 7-4 12	17 7-4 12	17 7-4 12
	..	17 7-4 12	17 7-4 12	17 7-4 12	17 7-4 12
	..	17 7-4 12	17 7-4 12	17 7-4 12	17 7-4 12
	..	17 7-4 12	17 7-4 12	17 7-4 12	17 7-4 12
	..	17 7-4 12	17 7-4 12	17 7-4 12	17 7-4 12
	..	17 7-4 12	17 7-4 12	17 7-4 12	17 7-4 12
	..	17 7-4 12	17 7-4 12	17 7-4 12	17 7-4 12
VI	A	9 7-5 12	9 7-5 12	9 7-5 12	9 7-5 12
	B	9 7-5 12	9 7-5 12	9 7-5 12	9 7-5 12
	C	9 7-5 12	9 7-5 12	9 7-5 12	9 7-5 12
	..	9 7-5 12	9 7-5 12	9 7-5 12	9 7-5 12
	..	9 7-5 12	9 7-5 12	9 7-5 12	9 7-5 12
	..	9 7-5 12	9 7-5 12	9 7-5 12	9 7-5 12
	..	9 7-5 12	9 7-5 12	9 7-5 12	9 7-5 12
	..	9 7-5 12	9 7-5 12	9 7-5 12	9 7-5 12
	..	9 7-5 12	9 7-5 12	9 7-5 12	9 7-5 12
	..	9 7-5 12	9 7-5 12	9 7-5 12	9 7-5 12

In comparing the chromosomal parameters in Table II it can be noted that all the *O. lacteum* biotypes with $2n = 12$ have a relative length of approximately 21 for chromosome I. The *O. lacteum* forma nov. (Sutherland) chromosome that has a relative length of 21 is thus probably chromosome I. Similarly chromosomes IV, V and VI occur in the karyotypes of all the *O. lacteum* biotypes including the forma nov. from Sutherland. The relative lengths of chromosomes II and III of the typical *O. lacteum* biotypes are approximately 19 and 18, respectively, and presumably they underwent centric union to produce the metacentric chromosome of the Sutherland forma which has a relative length of 37. The fact that chromosome III of the typical *O. lacteum* biotypes as well as the metacentric chromosome of the forma from Sutherland possess a satellite, is additional evidence in support of this assumption. Figure 4 illustrates how the presumed translocation could have taken place to give rise to the new karyotype.

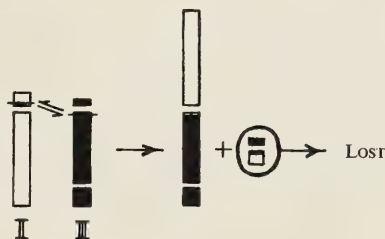


FIG. 4.

Diagram to illustrate how, by centric fusion, chromosome II and III of the primitive *Ornithogalum* complement gave rise to the long metacentric chromosome with a relative length of 37, in the karyotype of the *O. lacteum* forma nov. from Sutherland.

Column A of Table II gives an indication as to the proportional contribution of the different chromosomes to the whole complement. According to Swanson (1957) this is an important criterion in estimating the evolutionary position of a taxon, since a decrease in the chromosome number is correlated with evolutionary progress in many plant species.

Column B provides a measure of the symmetry of the chromosomes. The larger the figure in the column, the more asymmetrical are the chromosomes. The symmetry of the different chromosomes of one karyotype can be compared by reading the table horizontally while those of the different species and localities can be compared by reading the table vertically.

Column C gives the ratio of the short arm to the total length of the chromosome expressed as a percentage. Although there is an algebraic relationship between B and C, both are given, because C gives additional information about chromosome symmetry.

In the case of *O. thyrsoides*, a statistical analysis was made of the biotypes from 24 localities. A two way table (Table III) was compiled and in it the mean lengths of the chromosomes are presented as single observations. The least significant difference (L.S.D.) was calculated for the means of the chromosomes as well as for the localities, and are given at the foot of the Table.

TABLE III.

CHROMOSOME LENGTHS OF DIFFERENT BIOTYPES OF *O. THYRSOIDES*, (MEASURED IN MILLIMETRES AFTER IT WAS DRAWN AT A MAGNIFICATION OF 3,800 $\times$)

Localities	Chromosomes						Mean
	I	II	III	IV	V	VI	
Bokbaai	20.3	19.8	19.0	18.5	18.4	8.6	17.4
Calvinia	20.9	20.9	19.4	18.2	18.1	9.6	17.9
Citrusdal	20.6	22.6	19.9	22.1	23.3	9.8	19.7
Clanwilliam ..	20.7	20.3	20.8	19.7	18.0	8.5	18.0
Darling	21.4	21.0	21.1	20.5	20.3	9.0	19.0
Durbanville ..	22.2	20.7	20.9	20.6	18.6	9.4	18.7
Elsenburg	18.4	18.3	18.1	17.5	15.8	7.9	16.0
Gordons Bay ..	20.1	20.0	19.5	18.2	18.1	9.2	17.5
Grahamstown ..	22.4	21.0	20.0	19.6	16.1	9.8	18.2
Jonkershoek ..	27.0	26.4	24.3	23.0	21.6	11.2	22.3
Kamieskroon ..	18.9	18.0	17.0	16.7	15.0	8.8	15.7
Kirstenbosch ..	19.5	19.0	20.5	18.9	18.8	9.1	17.6
Kruisfontein ..	22.6	22.4	21.3	21.1	18.4	9.3	19.2
Leipoldville ..	18.4	18.3	17.6	18.1	15.8	8.1	16.1
Malmesbury ..	23.0	22.9	22.2	19.9	19.3	8.8	19.4
Melkbosstrand ..	22.0	21.8	22.0	20.6	18.7	8.9	19.0
Paleisheuwel ..	18.3	16.5	15.4	16.7	14.3	7.2	14.7
Porterville .. .	22.2	22.0	21.9	21.7	21.1	9.5	19.7
Riviersonderend ..	21.4	21.2	21.0	19.9	19.8	10.0	18.9
Sandveld	21.3	20.4	20.4	19.6	18.6	9.1	18.2
Springbok	19.5	18.5	17.7	17.4	17.1	8.9	16.5
Stellenbosch ..	25.2	23.9	23.4	22.4	20.9	10.2	21.0
Swellendam .. .	23.1	22.7	23.9	21.9	21.1	9.9	20.4
Welgevallen .. .	25.6	24.1	21.7	21.4	19.7	12.8	20.9
Mean	21.6	20.9	20.4	19.8	18.5	9.3	

L.S.D. for chromosome means = .50 with $P = .05$

= .65 with $P = .01$

L.S.D. for locality means = 1.0 with $P = .05$

= 1.3 with $P = .01$

From these calculated data Table IV was drawn up which gives an analysis of variance between biotypes and between chromosomes within biotypes.

TABLE IV.
ANALYSIS OF VARIANCE OF CHROMOSOME LENGTHS OF DIFFERENT BIOTYPES OF *O. THYRSOIDES*

Components of Variation				Degrees of Freedom	Sum of Squares	Mean Squares	Variance Ratio (F)
Localities	..	..	..	23	426.16	18.53	24.06**
Chromosomes	..	..	..	5	2523.50	504.70	655.45**
Error	..	..	..	115	88.32	.77	—
Total	..	..	..	143	3037.9	—	—

**P 0.01

The difference between chromosomes I and II is 0.7, which is significant. Between II and III, and III and IV the difference is only significant at the $P = 0.05$ level, and between IV and V, and V and VI the differences are again significant.

Chromosomes II and III differ from one another by the fact that one bears a satellite. The latter was taken as chromosome III.

By these means it was possible to distinguish all six of the chromosomes.

This study furthermore revealed that no distinction could be made between the typical *O. thyrsoides* biotypes and the *O. thyrsoides* formas on the basis of karyological characteristics.

From a comparison of the means and standard deviations which were calculated from Table I, it is also evident that on karyotypical grounds alone no distinction can be made between *O. thyrsoides*, *O. lacteum* and *O. conicum*.

DISCUSSION AND CONCLUSIONS

Like all systematic characters, the karyotype is subject to variation. In general, a particular karyotype can be designated as representative of the species, and in some instances even of the genus. In certain genera of the *Liliaceae* and in the North American species of *Tradescantia* the same karyotype is representative of an assemblage of species, while in *Crepis*, *Drosophila*, the lizards and the grasshoppers, the number, form and size of chromosomes vary widely from species to species (cf. Swanson, 1957).

The species of *Ornithogalum* in the present investigation showed a considerable degree of karyotypic resemblance. The statistical analyses indicate that no distinction can be drawn between biotypes and formas of *O. thyrsoides*, or

between different species on the basis of karyological differences. This is in good agreement with the findings of Stebbins (1950) for the family *Liliaceae*, of which *Ornithogalum* constitutes a genus.

Further information regarding crossability, meiotic studies of hybrids, and the primitiveness or specialisation of morphologic characters, etc., are needed for a sound natural classification of the species.

With a few rare exceptions such as the *O. lacteum* forma nov. from Sutherland, and the triploid and tetraploid plants in otherwise diploid populations, all plants of the species and biotypes investigated had a chromosome number of 12. This is in agreement with the results of Pienaar (1963).

According to de Wet (1957), most of the chromosomes of *Ornithogalum* are metacentric, while the current study and that of Pienaar (1963) revealed that all the chromosomes are acrocentric, that is, have subterminally located centromeres. The chromosome numbers for some species listed by de Wet (1957), are also at variance with the results of Pienaar (1963).

Swanson (1957) is of the opinion that there is no direct connection in either the plant or the animal kingdom between basic number and phylogenetic position of families and higher ranks of classification. Though differences may be apparent between phyletic groups, no obvious trends can be correlated with primitiveness or with specialisation unless such correlations are made within the much narrower limits of family and particularly genus. At this level, a general tendency towards reduction in basic number is an indication of specialisation. The members of the family or genus with the lowest chromosome numbers can, therefore, often be considered more specialised.

Aneuploidy and polyploidy undoubtedly play a very important role in the evolution of the karyotype. According to Stebbins (1950) polyploidy is more common in the plant kingdom. The older more primitive members have lower chromosome numbers.

Pienaar (1963) pointed out that polyploidy played no part in the origin and evolution of the *miniaturum-maculatum* group of species in Leighton's (1945) taxonomic list of the *Ornithogalum* species.

When only changes in basic number are involved, and the possibility of polyploidy definitely excluded, the major problem is one of determining the mechanisms leading to such change. A theoretical means of accomplishing this end was suggested by Darlington (1937). The scheme proposed was that gain or loss of chromosomes from a basic set involves gain or loss of centromeres, and that the success of such change depends upon whether the chromatin adjacent to the centromere is inert (heterochromatin) or active (euchromatin). Reciprocal translocations, with at least one of the breakage points being in inert regions, can transfer euchromatin to a centromere carrying only inert

chromatin. (A B-type chromosome would serve this purpose). In this manner a chromosome can be added. Euchromatin can also be transferred from one chromosome to another, with loss of the depleted and now inert centric chromosomal regions, thereby reducing the number.

The latter mechanism is thought to have taken place in the case of the *O. lacteum* forma nov. from Sutherland with $2n = 10$. From a comparison of the arm lengths of all the *O. lacteum* biotypes it could be concluded that during the evolution of the Sutherland forma a translocation took place between chromosomes II and III of the normal basic set, resulting in a long metacentric chromosome and a short fragmented chromosome, which was lost (Fig. 4).

Aneuploid alteration was carefully studied in the genus *Crepis* (Babcock and Cameron, 1934; Babcock, Stebbins and Jenkins, 1937, 1942; Babcock and Jenkins, 1943; Babcock, 1942, 1947) and it was concluded that the more primitive species have a higher basic number. The evolutionary trend has thus been towards a reduction in chromosome number.

Whether these trends also apply to the tribus *Scilleae* to which *Ornithogalum* belongs, cannot be confirmed at this stage. The basic chromosome number of *Ornithogalum* varies from $x = 3$ to $x = 9$ (Pienaar, 1963). Two closely related genera namely *Scilla* and *Urginea* possess a basic complement of $x = 4$ up to $x = 11$, and $x = 5$ respectively. It is thought that the primitive haploid number of *Ornithogalum* is $x = 6$ and that both a reduction and an increase had taken place during the course of evolution.

Together with changes in chromosome number, changes in the relative form and size of chromosomes are of considerable importance in a study of the karyotype.

Levitsky (1931b) made a very intensive study of the karyotypes of the family *Ranunculaceae* and concluded that a symmetrical karyotype is more primitive, and that asymmetry is correlated with specialisation. According to Stebbins (1950) a karyotype is symmetrical when the chromosomes are of approximately equal length and metacentric or submetacentric. Asymmetrical karyotypes consist of chromosomes with subterminal centromeres or large differences in lengths, or both.

The investigations of Babcock, Stebbins and Jenkins (1937), Babcock and Jenkins (1943), and Babcock (1947) in the genus *Cichoreae* of the family *Compositae*, confirmed Levitsky's hypothesis. The morphologically unspecialised species all possess symmetrical karyotypes. Stebbins (1950) believed that the family *Liliaceae* has the greatest diversity of karyotypes found within

one family. The more asymmetrical examples are found among the more specialised species.

From the present study and that of Pienaar (1963) it is evident that the South African species belonging to the genus *Ornithogalum* also possess karyotypes composed only of acrocentric chromosomes, many of which are markedly different in length.

In respect to these findings it appears that *Ornithogalum* must be fairly specialised, due to its reasonably asymmetrical karyotypes.

On these grounds it might be argued that the Sutherland biotype of *O. lacteum* forma nov., which possesses the metacentric chromosome, is a relic with a more primitive karyotype. However, this biotype is adapted to the more severe and specialised environmental conditions of Namaqualand, whereas most of the typical biotypes of *O. lacteum* grow in the milder regions of the South Western Cape. Its growth habit also appears to be slightly more specialised. Therefore this biotype is probably not a primitive relic, but must be regarded as a specialised form of *O. lacteum*, or another morphologically similar species. Its karyotype of $2n = 10$, although it mimics a more primitive condition, may consequently also be regarded as more evolved.

The tendency towards a symmetrical karyotype in the Sutherland forma of *O. lacteum* must be considered exceptional in *Ornithogalum*, because according to Swanson (1957) the presumed centric union which led to the formation of the metacentric chromosome is a rare event in higher plants although a rather common mechanism in karyotypic evolution in animals.

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